

## Sequence of a cDNA coding for human glutathione peroxidase confirms TGA encodes active site selenocysteine

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Selenium dependent glutathione peroxidase (1) is a nuclear encoded cytosolic and mitochondrial enzyme which maintains the integrity of DNA and lipids as well as reducing levels of endogenous hydrogen peroxide as follows:



where ROOH represents peroxidized DNA (2), lipid hydroperoxides, membrane-associated phospholipid hydroperoxides or hydrogen peroxide. We have isolated a cDNA coding the human enzyme from a kidney library in  $\lambda$ gt10 by cross-hybridization with a bovine cDNA (3); 24 of 5300 clones hybridized with the probe. The active site selenocysteine residue (-CH<sub>2</sub>SeH) at position 47 (i.e. SeC) is encoded by the nonsense codon, TGA, as is similarly observed in the mouse gene (4). Interestingly evidence suggests that the selenium atom is incorporated cotranslationally (5) rather than via a posttranslational modification step. This clone possesses 5 bp of the 5'-untranslated region, the 603 bp coding region, 223 bp of the 3'-untranslated region and a canonical polyadenylation signal, AATAAA, upstream of the polyA tract. The amino acid sequence reveals the protein possesses approximately 87% and 85% homology with preprocessed bovine (3) and mouse enzymes, respectively.

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      1      10      20      30      40      50      60      70      80      90      100      110      120      130      140      150      160      170      180      190      200      201
Met Cys Ala Ala Arg Leu Ala Ala Ala Gln Ser Val Tyr Ala Phe Ser Ala Arg Pro Leu Ala Gly Gly Glu Pro Val
GCCCC ATG TGT GGT GGT GGC CTA GCG GCG GCG GCG GCG GAG TCG GTG TAT GCG TTC TCG CCC GCG GCG CTC GCG GCG GCG GAG CCT GTG 89

Ser Leu Gly Ser Leu Arg Gly Lys Val Leu Leu Ile Glu Asn Val Ala Ser Leu [SeC] Gly Thr Thr Val Arg Asp Tyr Thr Gln Met Asn
ACC CTG GCG TCC CTG GCG GCG AAG GTA CTA CTT ATC GAG AAT GTG GCG TCC CTC [TGA] GCG ACC ACC GTC GCG GAG TAC ACC GAG ATG AAC 179

Glu Leu Cln Arg Arg Leu Gly Pro Arg Gly Leu Val Val Leu Gly Phe Pro Cys Asn Cln Phe Gly His Cln Glu Asn Ala Lys Asn Glu
GAG CTG CAG GCG GCG CTC GCA CCC GCG GCG CTG GTG CTC GCG TTC CCG TCG AAC CAG TTT GCG CAT CAG CAG AAC CCC AAG AAC GAA 269

Glu Ile Gln Asn Ser Leu Lys Tyr Val Arg Pro Gly Gly Gly Phe Glu Pro Asn Phe Met Leu Phe Glu Lys Cys Glu Val Asn Gly Ala
GAG ATT CAG AAT TCC CTC AAG TAC CTC GCG CCT GGT GGT GCG TTC GAG CCG AAC TTC ATG CTC TTC GAG AAG TCC CAG CTC AAC GGT GCG 359

Gly Ala His Pro Leu Phe Ala Phe Leu Arg Glu Ala Leu Pro Ala Pro Ser Asp Asp Ala Thr Ala Leu Met Thr Asp Pro Lys Leu Ile
GCG GCG CAG CCT CTC TTC CCG TTC CTG GCG GAG GCG CTC CCA GGT CCC AGC GAG CAG GCG ACC GCG CTT ATG ACC CAG GCG AAG CTC ATC 449

Thr Trp Ser Pro Val Cys Arg Asn Asp Val Ala Trp Asn Phe Glu Lys Phe Leu Val Gly Pro Asp Gly Val Pro Leu Arg Arg Tyr Ser
AGC TCG TCT GCG CTC TGT CCG AAC CAT GTT GCG TCG AAC TTT GAG AAG TTC CTC GTC GCG CCT CAG GGT GTG CCC CTA CCC AGG TAC ACC 539

Arg Arg Phe Cln Thr Ile Asp Ile Glu Pro Asp Ile Glu Ala Leu Leu Ser Gln Gly Pro Ser Cys Ala AM
GCG GCG TTC CAG ACC ATT CAG ATC CAG CCT CAG ATC GAA GCG CTC CTC TCT CAA GCG CCG AGG TGT GCG TAC GCGGCGGCTCTTACCGGCGCTGCT 635

TCCGAGTTGCACTGCTGCTGCTCTCTGCGGCGGCTTTTCATCTATGACGGCTGTTCTCTAAAGCTACGAGGAGCGAAGACCTGATCTTACAGAAAATACGACCTCGAGATCGCTGCTGCTG 755

TCTTCATCGCACTCTCTCGCCAGACCAAGCGCACTTTCGCGCACTAATAAAGTCCGCGCTCTGACCAAAAAA
Translated Mol. Weight = 21964.60

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## Nucleotide sequence of cDNA for rabbit glutathione peroxidase

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The cDNA coding rabbit glutathione peroxidase was isolated from liver cDNA library in lambda gtl1 by cross hybridization with the rat glutathione peroxidase cDNA which was cloned in this laboratory and reported elsewhere(1). The cDNA consisted of 600 bp of the coding region and the nucleotide sequence revealed that TGA, which is to be the stop codon in general, encoded seleno-cysteine(SeC) residue as was proved to be so with glutathione peroxidases of mouse (2), man(3) and rat(1). The amino acid sequence deduced from cDNA possesses 84, 84, 87 and 85% homology with rat, mouse, human, and bovine(4) enzymes, respectively.

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1 ATGTGTGGGCTCGTATGGCGGGCTGCCAGTCTGTGTACTCCTTCTCAGCGCACCCGCTGGCCGGCGGGGAG
M C A A R M A A A A Q S V Y S F S A H P L A G G E
76 CCCGTGAACCTGGGCTCCCTGCGGGGCAAGGTGCTGCTCATTGAGAATGTGGCGTGGCTGTGAGGCACTACGGTC
P V N L G S L R G K V L L I E N V A S L (SeC) G T T V
151 CGGGACTACACCCAGATGAACGAGCTGCAAGAGCGCCTCGGGCCCCGGGCCCTGGTGGTCTCGGCTTCCCGTGC
R D Y T Q M N E L Q E R L G P R A L V V L G F P C
226 AACGATTTGGGCATCAGGAGAACGCCAAGAATGAGGAGATTCTGAATTCCTCAAGTATGTCGGGCCTGGAGGC
N Q F G H Q E N A K N E E I L N S L K Y V R P G G
301 GGGTTCGAGCCCAACTTCATGCTCTTCCAGAATGCGAGGTGAACGGCGCCAAGGCCAGCCCGCTCTTCGCCTTC
G F E P N F M L F Q K C E V N G A K A S P L F A F
376 CTGCGGGAGGCCCTGCCGCCGCCAGCGACGCCCACTGCGCTCATGACCGACCCCAAGTTCATCACCTGGTGC
L R E A L P P P S D D P T A L M T D P K F I T W C
451 CCGGTGTGCCGTAACGACGTTTCTGGAGCTTCGAGAAGTCTGTTGGGCCCCGATGGTGTTCCTCGTGCAGG
P V C R N D V S W S F E K F L V G P D G V P V R R
526 TACAGCGCGCGCTTCCCAACCATCGACATCGAGCCCGACATCCAAGCCCTGCTGTCCAAGGGGTCTGGCGGTGCC
Y S R R F P T I D I E P D I Q A L L S K G S G G A
601 TAGgggccccctaccctggctgcttggcagtgccctgctgctctctgggggtttcatccatgafffgcttcccc
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676 cgaatacaaatggaggaacgcctgatgtccgggaacccccaggtggggcgtggtcotgtccatccc 742
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